

# The Effect of Sodium Chloride on the Growth and Variability of the Tadpole of the Frog.

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The investigations that have hitherto been carried out upon the effect of changes in their environment upon the variability of developing organisms are unfortunately not numerous. Indeed the researches of VERNON<sup>1)</sup> into the relation between variation in Echinoderm larvae and alterations in the temperature and the chemical composition of the medium in which they live stand almost alone.

And yet enquiries of this kind are of considerable interest and importance for they shed light not only on the phenomena of growth but also upon the causes of individual variations, while an examination of any changes in the value of correlations between organs that may occur may be expected to afford some indication of the extent to which each part is a self-differentiating system.

It may be worth while therefore to give here a brief account of a small series of experiments recently carried out upon the eggs and larvae of the common frog (*Rana temporaria*).

Newly laid eggs were taken from each of three batches (i. e. three pairs of parents) and placed in the followings solutions of sodium chloride in ordinary tap-water, namely .5%, .4%, .3%, .2%, .1%, while a sixth set were kept in tap-water as controls.

The eggs were placed, to the number of six or seven hundred (see Table I) in each jar, in jars of about four litres capacity in 3000 cc of water or salt-solution. The jars were lightly covered

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<sup>1)</sup> Phil. Trans. 186, B, 1895.

and all were immersed in a tank through which flowed a stream of running water, a constant temperature of about 9°C. being thus maintained.

At the end of a week or so, when the tadpoles were hatching out, all the eggs and larvae were killed and preserved in Formol and the experiment brought to an end.

In each jar the eggs and larvae fall obviously into two classes, 1) those which have hatched out and are either swimming about, feeding on the jelly or lying at the bottom: all these are normal in form; and 2) those which have failed to hatch out — having had their development arrested during segmentation or in the gastrula stage or during the closure of the medullary folds or possibly later — those which have hatched out but died and begun to degenerate, and those which whether hatched out or not are abnormal — with persistent yolk-plugs for example. Whether those which have failed to hatch out are alive or dead is not easy to tell unless disintegration has set in.

The accompanying table (Table I) gives the numbers, and percentages, for each of these two classes in each of the six media, and it will be seen at once that the »mortality« — if I may use the word to include all those cases which fall in the second class — is very high in the strongest solution, less but still high in the next two ( $\cdot 4\%$  and  $\cdot 3\%$ ), much less in  $\cdot 2\%$  and  $\cdot 1\%$ , while in the controls it is barely  $10\%$ .

Solutions of sodium chloride — even in these weak concentrations — are evidently injurious to the eggs and larvae.

The growth of the larvae is also affected.

Samples of those larvae that hatched out (class [1]) in  $\cdot 5\%$ ,  $\cdot 3\%$ ,  $\cdot 2\%$  and  $\cdot 1\%$ , have been measured in respect of their body-length (from the front of the head to the posterior border of the anus), tail-length (from the level of the posterior border of the anus to the tip of the tail) and total length (the sum of the two others).

The results are given in tenths of millimeters — each group is of two tenths of millimeters — in Tables II, III and IV.

In the case of each of these magnitudes the mean value ( $M$ ) is decreased by the action of sodium chloride and on the whole progressively decreased with increasing strength of the solution. The difference between the effect of the  $\cdot 2\%$  and that of the  $\cdot 3\%$  solution is, it is true, unimportant, but the effect of either of these is obviously less than that of the strongest solution employed.

Secondly, the variability is altered. This is not apparent if the standard deviations ( $\sigma$ ) alone be regarded, for, as the probable errors show, the differences between them are hardly significant, but if the coefficient of variability be considered (the standard deviation expressed as a percentage of the mean,  $\frac{\sigma}{M} \times 100$ ) then it is clear that the solution increases the variability and increasingly with the increase in its concentration, although again the effect of the .2% solution is practically the same as that of the .3%<sup>1)</sup>.

The alteration in the variability of the tail-length (Table III) is the most remarkable; in .5% the value is 37.76 as against 18.57 in the controls, just double, while its mean value in the former is barely more than half what it is in the latter.

It will be noticed at once that the tail-length is much more variable than the body-length in each solution.

It might be thought from this that there were as it were too independent growth centres for the body and the tail, and that the increase in size of the former was independent of the elongation of the latter.

This is however by no means the case as the values of the correlation coefficients calculated from the tables show (Tables V and VI).

In ordinary tap-water the coefficient ( $\rho$ ) is fairly high (.67) and its value in the strongest solution used (.5% NaCl) is practically the same (.59).

The solution therefore which interferes so seriously with the chances of hatching and living, with the growth and variability of the body and the tail of the tadpole but more especially of the latter, is powerless to alter the correlation between them. Whether the larvae developes well or ill the connection between these two parts of its structure remains unchanged.

It is hoped that in the future what promises to be a most interesting line of research will be carried out upon a larger scale.

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<sup>1)</sup> If the dead as well as the living had been rectored in the variability would of course have been larger still in the stronger solutions.

Table I.

	Dead, monstrous or unable to hatch		Hatched out and normal		Total Number
	Number	Per cent	Number	Per cent	
NaCl · 5%	486	74.88	163	25.12	649
NaCl · 4%	408	61.54	255	38.46	663
NaCl · 3%	470	60.33	309	39.67	779
NaCl · 2%	171	24.68	522	75.32	693
NaCl · 1%	162	21.23	601	78.77	763
Control	68	9.80	598	90.20	666

Table II.  
Length of Body.

Tenths of millimeters	Control	·2% NaCl	·3% NaCl	·5% NaCl
25—26				1
27—28				2
29—30			1	2
31—32		1	1	2
33—34	1	4	0	4
35—36	0	1	1	14
37—38	1	4	7	16
39—40	2	20	16	22
41—42	10	23	30	1
43—44	24	32	34	
45—46	34	15	10	
47—48	22		1	
49—50	4			
51—52	2			
Totals	100	100	101	64
$M$	45.02 ± .18	41.70 ± .20	41.76 ± .18	36.63 ± .28
$\sigma$	2.72 ± .13	3.00 ± .14	2.78 ± .13	3.34 ± .20
$\frac{\sigma}{M} \times 100$	6.04	7.19	6.66	9.12

Table III.  
Length of Tail.

Tenths of millimeters	Control	.2% NaCl	.3% NaCl	.5% NaCl
5—6			1	1
7—8		5	1	3
9—10		1	1	2
11—12	1	1	1	2
13—14	0	5	2	3
15—16	0	1	5	7
17—18	0	2	3	1
19—20	1	3	4	4
21—22	0	8	2	2
23—24	3	4	7	5
25—26	2	10	6	11
27—28	0	7	4	4
29—30	2	12	8	6
31—32	2	12	6	1
33—34	6	11	13	2
35—36	2	6	6	5
37—38	8	5	7	2
39—40	8	2	7	3
41—42	17	2	9	
43—44	10	2	5	
45—46	10	1	3	
47—48	8			
49—50	17			
51—52	2			
53—54	1			
Totals	100	100	101	64
$M$	$41.36 \pm .52$	$27.48 \pm .58$	$30.50 \pm .64$	$23.62 \pm .75$
$\sigma$	$7.68 \pm .37$	$8.68 \pm .41$	$9.50 \pm .45$	$8.92 \pm .53$
$\frac{\sigma}{M} \times 100$	18.57	31.59	31.15	37.76

Table IV.  
Total Length.

Tenths of millimeters	Control	.2% NaCl	.3% NaCl	.5% NaCl
31-32				1
33-34				1
35-36			1	1
37-38		1	1	0
39-40		2	0	2
41-42		1	0	2
43-44		0	0	1
45-46		2	0	0
47-48		0	0	1
49-50		2	1	2
51-52	1	3	1	2
53-54	0	1	3	2
55-56	0	1	3	5
57-58	1	1	2	3
59-60	1	2	4	7
61-62	0	4	3	4
63-64	0	6	3	8
65-66	4	5	6	4
67-68	1	7	5	2
69-70	1	7	3	4
71-72	0	15	5	2
73-74	1	13	10	3
75-76	3	7	9	4
77-78	4	6	10	1
79-80	4	4	7	2
81-82	7	3	5	
83-84	6	4	8	
85-86	14	1	8	
87-88	9	2	1	
89-90	6		1	
91-92	8		1	
93-94	6			
95-96	10			
97-98	10			
99-100	1			
101-102	1			
103-104	1			
Totals	100	100	101	64
$M$	$86.16 \pm .66$	$68.80 \pm .72$	$72.28 \pm .74$	$60.32 \pm .95$
$\sigma$	$9.88 \pm .47$	$10.62 \pm .51$	$10.88 \pm .52$	$11.30 \pm .67$
$\frac{\sigma}{M} \times 100$	11.47	15.44	15.05	18.73

Table V.  
Correlation between Tail Length and Body Length (Controls).

Tail	Body										
	51-52	49-50	47-48	45-46	43-44	41-42	39-40	37-38	35-36	33-34	
53-54	1										1
51-52			1	1							2
49-50	1	3	6	6	1						17
47-48		1	4	3							8
45-46			4	4	1	1					10
43-44			2	4	2	2					10
41-42			3	5	8	1					17
39-40			1	5	1	1					8
37-38				3	4	1					8
35-36				1	1						2
33-34			1	1	2	2					6
31-32				1	1						2
29-30					1		1				2
27-28											0
25-26					1					1	2
23-24					1	2					3
21-22											0
19-20								1			1
17-18											0
15-16											0
13-14											0
11-12							1				1
	2	4	22	34	24	10	2	1	0	1	100

$$r = .67 \pm .05.$$

Table VI.

Correlation between Tail Length and Body Length (.5% NaCl).

Tail	Body								
	41—42	39—40	37—38	35—36	33—34	31—32	29—30	27—28	25—26
39—40		2		1					3
37—38		1	1						2
35—36		4			1				5
33—34	1	1							2
31—32		1							1
29—30		3	1	2					6
27—28		1	1	1	1				4
25—26		4	3	3	1				11
23—24		1	2	2					5
21—22			1	1					2
19—20		1	2	1					4
17—18			1						1
15—16		3	3	1					7
13—14			1	1		1			3
11—12				1		1			2
9—10							2		2
7—8					1			1	3
5—6								1	1
	1	22	16	14	4	2	2	2	1
									64

$$r = .59 \pm .06.$$